

EFFECT OF DORMANCY ON THE EMERGENCE OF Tabernaemontana heterophylla SEEDLINGS IN DIFFERENT SUBSTRATES

Romário de Mesquita Pinheiro^{2*®}, Henrique Pereira de Carvalho^{3®}, Ricardo de Araújo Marques^{4®}, Evandro José Linhares Ferreira^{2®}, Gizele Ingrid Gadotti^{5®}, Quétila Souza Barros^{2®}, Andrea Alechandre da Rocha^{6®}, and Erica Karolina Barros de Oliveira^{7®}

1 Received on 22.11.2023 accepted for publication on 15.03.2024.

2 Instituto Nacional de Pesquisas da Amazônia, Rio Branco, Acre - Brasil. E-mail: <romario.ufacpz@hotmail.com>, <evandroferreira@ hotmail.com> and <quetilabarros@gmail.com>.

3 Universidade Federal do Acre, Graduando em Engenharia Agronômica, Rio Branco, Acre - Brasil. E-mail: https://www.engenharia.com br>.

4 Universidade Federal do Acre, Centro de Ciências da Saúde e do Desporto, Rio Branco, Acre - Brasil. E-mail: <ricardmarques@yahoo.com. br>.

5 Universidade Federal de Pelotas, Centro de Engenharias, Pelotas, Rio Grande do Sul - Brasil. E-mail: <gigadotti@hotmail.com>.

6 Universidade Federal do Acre, Centro de Ciências Biológicas e da Natureza, Rio Branco, Acre - Brasil. E-mail: .

7 Universidade Federal do Acre, Programa de Pós-graduação em Ciências Florestais, Rio Branco, Acre - Brasil. E-mail: <karolina.czs@gmail. com>.

*Corresponding author.

ABSTRACT

The composition and dynamics of plant communities are determined by the survival, dispersal and persistence of seeds in the soil, some of the fundamental aspects for understanding plant biology, among which seed dormancy is of particular importance. The aim of this work was to find out whether seed dormancy is affected by certain substrates in order to improve the production of *Tabernaemontana heterophylla* seedlings. Treatments were carried out with different substrates (T1. Sand, T2. Vegetable clay, T3. Vegetable clay + sand (1:1 ratio), T4. Vermiculite, T5. Cattle manure, T6. Sawdust and T7. Organic compost) in 7 x 4 factorial schemes, the results of which were subjected to an analysis of variance and a comparison of means with the Tukey test at a probability of 5%. The seeds with visible germination structure (hypocotyl) showed an apical meristem in a dormant stage. The number of normal seedlings did not exceed 25% of the germinated seeds in all treatments, with a high proportion of nonviable seeds observed. In the absence of technical information on overcome dormancy, it is important to use strategies that stimulate germination and subsequent seedling growth. The slow germination combined with the presence of dormancy in the hypocotyledon were factors that negatively affected the propagation of the species studied.

Keywords: Forest species; Germination; Forest seeds.

How to cite:

Pinheiro, R. de M., Carvalho, H. P. de, Marques, R. de A., Ferreira, E. J. L., Gadotti, G. I., Barros, Q. S., Rocha, A. A. da, & Oliveira, E. K. B. de. (2024). Effect of dormancy on the emergence of *Tabernaemontana heterophylla* seedlings in different substrates. *Revista Árvore, 48*(1). https://doi.org/10.53661/1806-9088202448263743. Retrieved from: https://www.revistaarvore.ufv.br/rarv/article/view/263743









EFEITO DA DORMÊNCIA NA EMERGÊNCIA DE PLÂNTULAS DE Tabernaemontana heterophylla EM DIFERENTES SUBSTRATOS

RESUMO – A composição e a dinâmica das comunidades vegetais são determinadas pela sobrevivência, dispersão e persistência das sementes no solo, alguns dos aspectos fundamentais para a compreensão da biologia vegetal, entre os quais se destaca a dormência das sementes. O objetivo deste trabalho foi verificar se a dormência das sementes é afetada por determinados substratos, a fim de melhorar a produção de mudas de Tabernaemontana heterophylla. Os tratamentos foram realizados com diferentes substratos (T1. Areia, T2. Argila vegetal, T3. Argila vegetal + areia (proporção 1:1), T4. Vermiculita, T5. Estrume de gado, T6. Serragem e T7. Composto orgânico) em esquemas fatoriais 7 x 4, cujos resultados foram submetidos a uma análise de variância e a uma comparação de médias com o teste de Tukey a uma probabilidade de 5%. As sementes com estrutura germinativa visível (hipocótilo) apresentaram meristema apical em estado de dormência. O número de plântulas normais não ultrapassou 25% das sementes germinadas em todos os tratamentos, tendo sido observada uma elevada proporção de sementes inviáveis. Na ausência de informações técnicas sobre a superação da dormência, é importante utilizar estratégias que estimulem a germinação e o posterior crescimento das plântulas. A lenta germinação aliada à presença de dormência no hipocótilo foram fatores que afetaram negativamente a propagação da espécie estudada.

Palavras-Chave: Espécie florestal; Germinação; Sementes florestais.

1. INTRODUCTION

The growing need for seeds and seedlings studies is directly related to the increasing use of native tree species in initiatives aimed at forest restoration, environmental recovery, conservation of natural resources and medicinal applications. The plant popularly known as "Grão-de-Galo" (*Tabernaemontana* *heterophylla* Vahl) belongs to the Apocynaceae family. It is native to the tropical region of South America, including countries such as Brazil, Colombia, Venezuela and Guyana (Kew, 2023). The use of this species in medicine has gained importance and is increasingly being researched. The species has methanolic extracts in its woody stem. However, the main target for study and extraction are the indolic alkaloids, mainly the iboga type (Wolter Filho et al., 1983).

Tabernaemontana is a genus used in traditional medicine for its antidepressant and anti-addictive effects on the central nervous system (Cloutier-Gill et al., 2016; González-Trujano et al., 2022). Although there is a large amount of medicinal information about this genus, there is still a lack of knowledge about the possibilities of propagating this species, especially in terms of overcoming seed dormancy. Studies at the plant production level on the characteristics of seed dormancy are important to understand and determine the most appropriate propagation methods for a given species (Samarasinghe et al., 2022).

To be successful in the utilization of native forest species, it is essential to understand and identify their germination characteristics, develop methods to optimize seed germination process and find the ideal substrate, which is crucial environmental factor for the success of the germination process. This not only helps to gain important knowledge for the production of seedlings of these species, but is also crucial for promoting the cultivation of species that have great potential in ecological, economic and medicinal terms.

Seed germination and seedling emergence are strongly influenced by a type of dormancy, in addition to the type of sowing substrates, which are crucial for seedling quality (Han et al., 2018, Sousa et al., 2023). Among the numerous elements that affect seedling production, substrate composition not only influences seed germination but also plays a crucial role in seedling support and development by providing nutrients, water and oxygen (Silva et al., 2023) and promoting higher photosynthetic efficiency (Afonso et al., 2020). The choice of substrate must be adapted to the specific needs of the seed in terms of its size and shape.

It is important to explore the use of new



alternative materials as substrates in order to evaluate their potential for seedling production while trying to reduce production costs. It is also crucial to identify the types of seed dormancy, as these are important adaptive traits that help ensure that seed germination only occurs under favorable conditions for good establishment and successful growth in cultivated areas or in the natural environment. Seed dormancy can be categorized into different classes: physiological, physical, combined (physiological and physical), morphological morphophysiological and (Baskin and Baskin, 2021). Information on dormancy and germination of native species seeds is important to diversify the use of species in restoration programs (Pedrini and Dixon, 2020).

The production of seedling from seeds is the most important means of propagation of *Tabernaemontana heterophylla*. Therefore, knowledge of seed dormancy and germination is useful for the pharmaceutical industry and for conservation of the species. In view of this, the present study was conducted to determine the role of seed dormancy in the emergence of seedlings in different substrates to improve the production of *Tabernaemontana heterophylla* seedlings.

2. MATERIAL AND METHODS

The work was carried out in the forest seed laboratory and in the seedling greenhouse of the Parque Zoobotânico at the Federal University of Acre, in the municipality of Rio Branco, Acre. Freshly harvested *Tabernaemontana heterophylla* seeds from a forest fragment near the city of Rio Branco, Acre, were used.

The seeds were harvested directly from the mother plant, from fruits that were at the stage of physiological maturity for dispersal, with the visible opening of the dried fruit showing its orange-colored inner part with numerous seeds still partially covered by the aril. The seeds were packed in a cooler and brought to the laboratory. The aril was removed by rubbing and washing the seeds with water in a sieve. To remove excess water, the seeds were placed on paper to dry naturally.

As we do not know any specific propagation methods for this species, we first decided

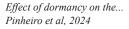
to test germination in the laboratory using methods to overcome dormancy. The aim was to investigate the possible existence of integumentary dormancy and thus determine the most effective method to overcome it. The pre-tests used were: scarification with No. 80 sandpaper and soaking in water at room temperature for 24 hours; trimming and soaking in water at room temperature for 24 hours; scarification with No. 80 sandpaper; trimming; immersion in water heated to 80 °C for five minutes and control - no treatment.

The seeds were scarified at the point opposite the hilum by rubbing the seed with sandpaper until part of the tegument broke off, and then soaked for 24 hours or not. Seed trimming consisted of a cut on the side near the hilum, removing only the tegument, followed by soaking for 24 hours or not. Soaking was carried out in water heated to 80 °C for five minutes (H₂O 80 °C/ 5'), followed by removal of the seeds from the water and cooling to room temperature.

Following these procedures, it was found that the method of trimming the seed without immersion in water resulted in a higher percentage of germinated seeds from a biological point of view when there were visible signs of germination (swelling of the endosperm exposing the integument or the beginning of radicle protrusion). This method was therefore used to conduct the study with the different substrates, which were as follows: T1. Sand (sieved), T2. Vegetable clay (collected from the forest), T3. Vegetable clay + sand (1:1 ratio), T4. Vermiculite (sterilized), T5. Cattle manure (tanned), T6. Sawdust (tanned) and T7. Organic compost (decomposed plant waste).

For the emergence test, which was carried out in the greenhouse, the percentage, average time and average speed of seedling emergence were determined. Each treatment consisted of 100 seeds, divided into four replicates of 25 seeds each, sown on the different substrates in plastic trays measuring 53 x 38.3 x 8.4 cm. During the experiment, the natural temperature in the greenhouse fluctuated between 30 and 37 °C. The germinated seeds were counted daily, counting the seedlings that showed the emission of the hypocotyl and the development of the first pair of cotyledonary leaves.

Calculations of percentage emergence,





average time, speed and frequency of seedlings were carried out according to the equations developed by Labouriau and Valadares (1976).

- Percentage of emergence (%E):

$$E = \left(\frac{N}{A}\right) \cdot 100 \qquad (Eq.1)$$

where: E= seedling emergence percentage; N= number of emerged seedlings; A= total number of seeds placed to germinate.

- Average emergence time (AET):

$$t = \frac{\left(\sum_{i=1}^{k} ni \cdot ti\right)}{\sum_{i=1}^{k} ni}$$
(Eq.2)

where: t = average incubation time; ni = number of emerged seedlings per day; ti = incubation time (days).

- Average speed of emergence (ASE):

$$\mathbf{v} = \frac{1}{t} \tag{Eq.3}$$

where: V = average speed of emergence (days); t = average time of emergence.

- Emergence frequency:

$$Fr = \frac{ni}{\sum_{i=1}^{k} ni}$$
(Eq.4)

where: Fr = relative frequency of emergence; ni = number of seedlings emerged per day; Σ ni = total number of seeds germinated.

At the end of the experiment, the percentage of non-viable (hard seeds) and viable seeds (green seeds) was counted. No formal tests were carried out, but it was observed that all the seeds that germinated and were viable had a green color, which could be seen when breaking open the tegument, a characteristic of the species. In contrast, the non-viable seeds were hardened and deteriorated, turned whitish in color due to decomposition.

The experimental design was completely randomized and consisted of a 7×4 factorial scheme (seven treatments and four replicates). The data obtained were subjected to analysis

of variance. If F-test was significant, the means were compared with the Tukey test at 5% probability. The software used for the analysis was WinStat (Machado et al., 2001).

3. RESULTS

After performing the pre-test to overcome the seed tegument dormancy by trimming the lateral region near the hilum, it was found that this species had an external protuberance of the embryo that partially exposed the endosperm from which the radicle or hypocotyl emerged (Figure 1). The experiment was evaluated for 125 days and showed a high rate of seeds that failed to germinate. Over time, the seeds with a visible hypocotyl structure exhibited a dormant apical meristem, indicating a lack of visible growth combined with reduced metabolism. It can therefore be assumed that the species has a hypocotyl-apical dormancy. Despite a significant delay, the seedlings eventually resumed growth.

From the information shown in figure 1e, it appears that the species germinates epigeically, with the seedling being phanerocotyledonous with leaf-like cotyledons. This indicates that dormancy of the seedling is not associated with growth of the primary root and/or hypocotyl. Rather, it appears to be related to apical dormancy, as evidenced by the delay in the appearance (development) of the eophilus(es) - the first leaves - and the elongation of the epicotyl. It is important to note that there was no delay in the elongation of the root and hypocotyl after protrusion, suggesting that this is not a characteristic of low vigor seeds, but is due to dormancy. The germination rate and formation of normal seedlings was very low, averaging <25 % of seeds (Figure 2), resulting in uneven germination, even with the use of methods to overcome tegument dormancy.

Seedling emergence in the different substrates was evaluated considering the applied treatment to overcome dormancy (seed trimming), which gave the best germination performance. Seedling emergence was slow and irregular in all treatments. The average scores for the vegetable clay substrate treatment (T2), with 21% seedling emergence, was higher than the other treatments, indicating a better result for germinated seed, but there was no difference in the average speed of emergence between the treatments evaluated (Figure 2).



Percent seedling emergence for treatment T2 was statistically different from the substrates with Vermiculite (T4), Cattle manure (T5), Sawdust (T6) and Organic compost (T7) (p-value = 0.0009, significance at 0.05%). The higher the seedling emergence, the lower the average speed, indicating that ASE is reduced by the amount of non-germinated seeds that will form a normal, vigorous seedling.

The average emergence time was calculated from the time the experiment was set up until the last day of evaluation. In this study, the evaluation lasted 124 days (Figure 3). Above-ground shoot development was prolonged in all treatments, regardless of the substrate used. In the substrates sand (T1) and vermiculite (T4), emergence began after 38 days and lasted a further 81 and 79 days respectively. In vegetable clay (T2), emergence began after 70 days and lasted a further 49 days. In the substrate made up of clay + sand (T3) seedling emergence began on day 34, and continued for a further 85 days. In cattle manure (T5), the first seedling emergence was observed after 40 days, but it was aborted shortly afterwards when it was found that more than 50% of the seeds were not viable (Table 1).

In the substrate with sawdust (T6), emergence was observed 114 days after sowing and continued for a further 10 days. In the organic compound (T7) emergence began after 56 days and was aborted shortly thereafter due to the high proportion of non-viable seeds. The average total time for seedling emergence (considering all treatments) was 87 days.

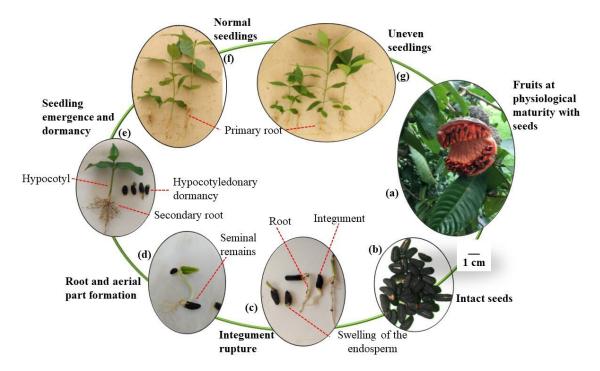


Figure 1 - Morphological aspects of seed germination and seedling establishment of *Tabernaemontana heterophylla*: (a) Physiologically mature fruit attached to the mother plant ready for dispersal of its seeds (b) processed seeds, (c) start of root protrusion and/or hypocotyl, as well as the appearance of the endosperm after overcoming dormancy by seed trimming (7 to 10 days), (d) development of the roots and aerial part of the seedling (35 days), (e) emergence of seedlings and dormancy in the apical meristem (75 days), (f) normal seedlings and (g) characterization of the uniformity of the seedlings caused by dormancy (124 days).

Figura 1 - Aspectos morfológicos da germinação e estabelecimento de plântulas em sementes de *Tabernaemontana heterophylla*: (a) Fruto em maturidade fisiológica ligado a planta mãe pronto para dispersão de suas sementes (b) sementes beneficiadas, (c) início da protrusão radicular e/ou hipocótilo, além do aparecimento do endosperma pós a superação de dormência por desponte (7 a 10 dias), (d) desenvolvimento da raízes e parte aérea da plântula (35 dias), (e) emergência de plântulas e dormência no meristema apical (75 dias), (f) plântulas normais e (g) caracterização da uniformidade das plântula ocasionado por dormência (124 dias).



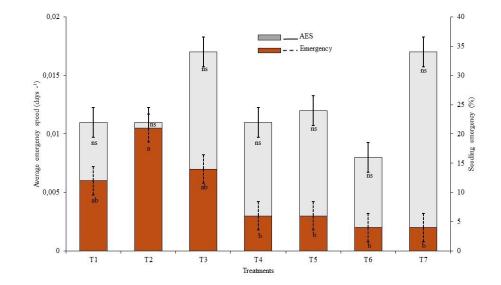


Figure 2 - Percentage of emergence and average seedling speed in a stacked graph on the response variables. T1 = Sand; T2 = Vegetable soil; T3 = Vegetable soil + sand; T4 = Vermiculite; T5 = Cattle manure; T6 = Sawdust; and T7 = Organic compost. Mean values followed by the same letters, comparing the mentioned treatments, did not show significant differences according to the Tukey test (P<0.05). *Represents the existence of a difference between the variables. Bars represent the standard error of the mean of four repetitions, and ns indicates non-significance.

Figura 2 - Percentual de emergência e velocidade média de emergência de plântulas em gráfico empilhado sobre as variáveis resposta. T1 = Areia; T2 = Barro vegetal; T3 = barro vegetal + areia; T4 = Vermiculita; T5 = Esterco bovino; T6 = Pó de serra e T7 = Composto orgânico. Os valores médios seguidos das mesmas letras, comparando os tratamentos citados, não apresentaram diferença significativa pelo teste de Tukey (P<0,05). *representa a existência de diferença entre as variáveis. Barras representam o erro padrão da média de quatro repetições e ns não significativo.

When treatments were individualized, the following results were observed: T1 (97 days), T2 (88 days), T3 (66 days), T4 (88 days), T5 (86 days), T6 (124 days) and T7 (57 days), all with a low germination rate. It should be noted that the evaluation of treatments T5 and T7, which had low values for emergence time, was terminated once it was determined that more than 50% of the seeds were not viable (Table 1).

The average emergence time values showed significant differences between the evaluated treatments (Figure 3), especially between T6 and T7.

The seeds of *Tabernaemontana heterophylla* exhibit recalcitrant behavior (a condition observed during in the storage of a batch of seeds that lost viability within a few days), which, together with the dormancy observed in the seeds, hinders the development and establishment of seedlings.

The initial stage of seedling emergence varied between treatments, with no simultaneous germination. The distribution of the frequency of emergence showed polymodality in all treatments, with the polygonal line touching the horizontal axis more than once, indicating multiples peaks of seedling emergence (Figure 4), a condition that characterizes the germination process as slow and irregular.

Based on the daily distribution of germination frequency, the following observations could be made: there were intervals between germination peaks in all treatments there were spaced intervals of seedling emergence from one to the next (Figure 4), except for treatments T5 and T7, which were terminated prematurely due to the high proportion of non-viable seeds. The other treatments had a higher number of normal seedlings, but at irregular intervals, resulting



Table 1 - Characteristics of seeds during the experiment in terms of their germination aspects. SM = dead seeds, SD = hard seeds, and Emerg. = seeds that gave rise to a normal seedling.

Tabela 1 - Características das sementes durante a condução do experimento, quanto ao seu aspecto de germinação. SM = sementes mortas, SD = sementes duras e Emerg. = sementes que originaram uma plântula normal.

Treatments —	Seed aspects		
	SM	SD	Emerg.
		%	
T1. Sand	39c*	49b	12ns
T2. Vegetable soil	25d	54ab	21
T3. Vegetable soil + sand	75b	11d	14
T4. Vermiculite	92a	2e	6
T5. Cattle manure	58b	33c	6
T6. Sawdust	36c	60a	4
T7. Organic compost	80a	16d	4
Overall average	57	30	10

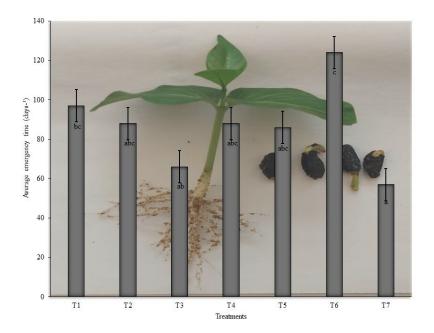


Figure 3 - Average emergence time as a function of pre-established treatments. T1 = Sand; T2 = Vegetable soil; T3 = Vegetable soil + sand; T4 = Vermiculite; T5 = Cattle manure; T6 = Sawdust; and T7 = Organic compost. Mean values followed by the same letters, comparing the mentioned treatments, did not show significant differences according to the Tukey test (P < 0.05). *represents the existence of a difference between the variables. Bars represent the standard error of the mean of four repetitions.

Figura 3 - Tempo médio de emergência em função dos tratamentos pré-estabelecidos. T1 = Areia; T2 = Barro vegetal; T3 = barro vegetal + areia; T4 = Vermiculita; T5 = Esterco bovino; T6 = Pó de serra e T7 = Composto orgânico. Os valores médios seguidos das mesmas letras, comparando os tratamentos citados, não apresentaram diferença significativa pelo teste de Tukey (P<0,05). *representa a existência de diferença entre as variáveis. Barras representam o erro padrão da média de quatro repetições.



in inconsistent seedlings establishment. The most significant peaks were observed in seedling emergence, considering those that germinated daily in all replicates of the treatments.

When the polygonal line is shifted to the right without reaching the horizontal axis, a series of daily germination events is observed (Figure 4). However, when the peaks represented by non-collinear lines touch or approach the horizontal axis after germination, uneven germination peaks occur (Figure 4). This results in no germination on some of the days observed in certain replicates.

4. DISCUSSION

The low seedling emergence in the substrates can be attributed to temperature conditions, water holding capacity, aeration and hormonal factors. This limitation may be exacerbated by the lack of comprehensive information on specific dormancy overcoming promote both techniques that seed germination and subsequent seedling growth of Tabernaemontana heterophylla. The results of this study indicate low seedling emergence, and none of the treatments significantly favored uniform performance, as the 50% or 100% mark of emerging seedlings was not reached.

Tabernaemontana heterophylla has recalcitrant seeds that favor a decrease in their physiological quality due to the long germination period and the high water content in their structures. This leads to a decrease in reserves and degradation of essential metabolites important for the germination process, conditions that cause the seeds to lose their viability, followed by deterioration (Cavalcante et al., 2023).

To preserve seeds in unfavorable environments that could jeopardize seedlings survival and establishment, plants have evolved a wide range of dormancy strategies (Nonogaki, 2019, Garcia et al., 2020). These strategies coordinate the germination and emergence of seedlings and ensure that they emerge at the right time for their survival. *Tabernaemontana heterophylla* has a dormancy manifested by a rigid integument and a dormant physiological component (hypocotyl in the dormant stage). Hypocotyledon apical dormancy can occur depending on the photoperiod and the occurrence of warm or hot temperatures. In this study, which was conducted in a greenhouse, there was no temperature control.

As global warming progresses, species evolves strategies for reproduction as they absorb seasonal information from the environment and pass it on their offspring (Pinheiro et al., 2021, Rosbakh et al., 2023). A study conducted out by Moraes et al. (2016) with *Tabernaemontana fuchsiaefolia* A. DC reports the analysis of the physiological quality of seeds exposed to high temperature stress through accelerated aging, resulting in a low percentage of seed germination.

Tabernaemontana heterophylla appears to form a seedling bank rather than a seed bank as part of its survival strategy. The emergence of the hypocotyl above the substrate cause the cotyledons (paracotyledons) to rise above the surface of the substrate and be released, characterizing a normal seedling.

In this study, the elongation of the embryo and the emergence of the radicle showed that integumentary dormancy was overcome, although it took longer for normal seedlings to form. In many tropical species with underdeveloped embryos, a physiological component of dormancy may also be present during seedling development (Gemma et al., 2023). Twig dormancy that does not allow branching has also been reported (Streubel et al., 2023). Zhang et al. (2022) observed the morphophysiological dormancy of epicotyl in seeds of *Paeonia ostii* (Paeoniaceae) when they investigated the seasonal regulation of temperature in germination phenology and found that emergence of radicles occurs only at warm temperatures.

The materials or compositions present in the substrates have different effects on seedling emergence, a crucial stage in the plant's development cycle. This stage is crucial for the proper establishment of plants in the field, as they are highly susceptible to environmental stresses (Porceddu et al., 2016). This effect manifests itself quantitatively in the production of biomass and, consequently, influences the initial growth of the seedlings. Rarely will a single substrate type be able to fulfill all the specific requirements of the species to be propagated (Peng et al., 2023). For this reason, materials that improve the physical properties are often added to the



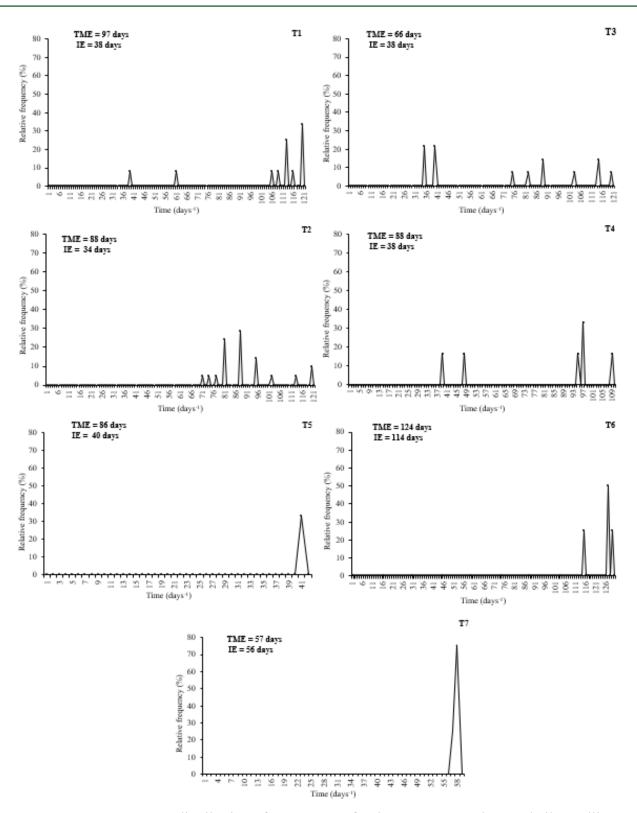


Figure 4 - Frequency distribution of emergence of *Tabernaemontana heterophylla* seedlings subjected to different substrates. T1 = Sand; T2 = Vegetable soil; T3 = Vegetable soil + sand; T4 = Vermiculite; T5 = Cattle manure; T6 = Sawdust; and T7 = Organic compost.

Figura 4 - Distribuição de frequência de emergência de plântulas *Tabernaemontana heterophylla* submetidas a diferentes substratos. T1 = Areia; T2 = Barro vegetal; T3 = Barro vegetal + Areia; T4 = Vermiculita; T5 = Esterco bovino; T6 = Pó de serra e T7 = Composto orgânico.



substrates. It can therefore be concluded that the low percentage of seedling emergence found in this study was not directly influenced by the evaluated substrates.

On the other hand, uniform substrate moisture must be maintained to ensure that the seeds receive the amount of water required for germination. An excess of water can accelerate decay and create conditions for the growth of microorganisms, while a lack of water can interrupt vital metabolic processes (Oliveira et al., 2015). The response variables analyzed, such as the average speed, average time and percentage of seedling emergence, revealed that *Tabernaemontana heterophylla* presents germination peaks that turn into a dormant phase after epicotyl emission, resulting in seedling non-uniformity.

Proper seedling production requires uniform establishment and development in terms of seedling size, and seed quality is critical to ensure this uniformity. On the other hand, it is important to understand the germination stages associated with the substrates, as the water retention capacity of the substrate together with the morphological characteristics of the seeds, can influence the percentage of germinated seeds (Silva et al., 2023).

Although the germination of seeds subjected to the method of overcoming dormancy did not vary significantly depending on the different substrates compared to the normal seedlings, it should be noted that biological development undergoes various changes during the germination process. These changes may indicate species-specific adaptations and responses to the environment, suggesting an influence of apical meristem dormancy on the initial development of the seedlings. This confirms that seed germination involves complex physiological processes that respond to environmental signals such as water potential, light, hormones and oxygen (Bano et al., 2021, Pinheiro et al., 2021).

Besides moisture, temperature is considered the most important limiting factor for seed germination (Malek et al., 2022). The right temperature plays a crucial role in the activation of metabolic and enzymatic processes that are essential for the start of embryonic development during germination. Temperature fluctuations can have a direct effect on the speed and uniformity of germination. Therefore, it is important to consider the ideal thermal conditions to promote an efficient germination process in different plant species (Garcia et al., 2020, Jhanji et al., 2024). The rate at which germination or the emission of the primary root occurs is subject to a variety of edaphoclimatic conditions that play a crucial role in regulating metabolism, water uptake and biochemical reactions (Bewley et al., 2013).

Although there was no significant variability in the germination of seeds subjected to the method of overcoming tegument dormancy in different substrates, as far as normal seedlings are concerned, it can be seen that biological development undergoes various changes during the germination process. These changes may indicate speciesspecific adaptations and responses to the environment, suggesting an influence of some type of dormancy on the initial development of the seedlings (Langa et al., 2024). Seed germination and performance vary between different plants, resulting in different types of dormancy (Onol and Yildirim, 2021). This variability can be influenced by a series of genetic, environmental and physiological factors, contributing to the complexity of the germination process in various species.

5. CONCLUSION

The results showed that the presence of hypocotyledonary dormancy and slow seed germination resulted in uneven seedling establishment, negatively affecting the propagation of *Tabernaemontana heterophylla*. Furthermore, it was found that the different substrates did not exert a significant influence on the germination process.

AUTHOR CONTRIBUTIONS

Romário de Mesquita Pinheiro: Conceptualization, Methodology, Validation, Investigation, Writing - Original Draft, Supervision; Henrique Pereira de Carvalho and Ricardo de Araújo Marques: Methodology, Validation, Investigation; Evandro José Linhares Ferreira: Conceptualization, Methodology, Validation, Investigation, Writing, english translation-Original Draft; Quétila de Souza Barros : Conceptualization, Methodology, Validation, Writing - Original Draft; Gizele Ingrid Gadotti: Conceptualization, Methodology,



Validation, Investigation, Writing - Original Draft ; Andrea Alexandre da Rocha and Erica Karolina Barros de Oliveira: Writing - Review & Editing, Visualization.

7. REFERENCES

Afonso MV, Paranhos JT, Tabaldi LA, Soriani HH, Saldanha, CW. *Tabernaemontana catharinensis* A. DC. seedling emergence and growth in different substrates. Floresta E Ambiente. 2020;27(2):e20170770. doi. https://doi.org/10.1590/2179-8087.077017.

Bano H, Rather RA, Bhat JI, Bhat TT, Azad H, Bhat SA, Bhat MA. Effect of presowing treatments using phytohormones and other dormancy breaking chemicals on seed germination of *Dioscorea deltoidea* Wall. Ex Griseb.: an endangered medicinal plant species of north western Himalaya. Ecology, Environment and Conservation. 2021;27:253-260.

Baskin JM, Baskin CC. The great diversity in kinds of seed dormancy: a revision of the Nikolaeva–Baskin classification system for primary seed dormancy. Seed Science Research. 2021;31:249-277. doi:10.1017/ S096025852100026X.

Bewley JD, Bradford KJ, Hilhorst HWM, Nonogaki H. Seeds: Physiology of Development, Germination and Dormancy. 3rd ed. New York: Springer; 2013. 392p. doi. 10.1007/978-1-4614-4693-4.

Cavalcante JA, Gadotti GI, Pinheiro R de M, Silva RNO da, Oliveira FK de, Moraes DM de. Vigor and anaerobic metabolism of soybean seeds evaluated by ethanol test. Journal Seed Science. 2023;45:e202345007. doi. https://doi.org/10.1590/2317-1545 v45263825.

Cloutier-Gill L, Wood E, Millar T, Ferris C, Socias ME. Remission of severe opioid use disorder with ibogaine: a case report. Journal of Psychoactive Drugs. 2016;48:214-217. doi. https://doi.org/10.1080/02791072.2016.1180 467.

Garcia QS, Barreto LC, Bicalho EM. Environmental factors driving seed dormancy and germination in tropical ecosystems: A perspective from campo rupestre species. Environmental and Experimental Botany. 2020;178:104164. doi. https://doi.org/10.1 016/j.envexpbot.2020.104164. Gemma HL, Stevens AV, Guja LK, Sommerville KD, Worboys S, Crayn DM. Sunlight and red to far-red ratio impact germination of tropical montane cloud forest species. Australian Journal of Botany. 2023;71:326-339. doi. https://doi.org/10.1071/ BT22126.

González-Trujano ME, Krengel F, Reyes-Chilpa R, Villasana-Salazar B, González-Gómez JD, Santos-Valencia F, Urbina-Trejo E, Martínez A, Martínez-Vargas D. *Tabernaemontana arborea* and ibogaine induce paroxysmal EEG activity in freely moving mice: Involvement of serotonin 5-HT1A receptors. NeuroToxicology. 2022;89:79-91. doi. https://doi.org/10.1016/j. neuro.2022.01.002.

Han AR, Kim HJ, Jung JB, Park PS. Seed germination and initial seedling survival of the subalpine tree species, *Picea jezoensis*, on different forest floor substrates under elevated temperature. Forest Ecology and Management. 2018; 429:579-588. doi. https://doi.org/10.1016/j.foreco.2018.07.042.

Jhanji S, Goyal E, Chumber M, Kaur G. Exploring fine-tuning between phytohormones and ROS signaling cascade in regulation of seed dormancy, germination and seedling development. Plant Physiology and Biochemistry. 2024;207:108352. doi. https:// doi.org/10.1016/j.plaphy.2024.108352.

Kew. Royal Botanic Gardens. *Tabernaemontana heterophylla* Vahl. Disponível em > https://powo.science.kew. org/taxon/urn:lsid:ipni.org:names:82146-1#publications. Acesso em 15 de nov. 2023.

Labouriau LG, Valadares MEB. On the germination of seeds *Calotropis procera* (Ait.) Ait.f. Anais da Academia Brasileira de Ciências. 1976;48(2):263-284.

Langa S, Magwaza LS, Mditshwa A, Tesfay SZ. Seed dormancy and germination responses of *Cannabis landraces* to various pre-treatments. South African Journal of Botany. 2024;165:91-100. https://doi.org/10.1016/j. sajb.2023.12.021.

Machado AA, Conceição AR, Silva JGCE, Campari CAP, Júnior PS, Porenstein D, Krolow RALA, Gonsales AD, Junior JCV. WinStat. Sistemas de análises estatísticas para Windows. Versão 2.11. UFPEL–Universidade Federal de Pelotas, NIA – Núcleo de Informática Aplicada. 2001.



Malek M, Ghaderi-Far F, Torabi B, Sadeghipour HR. Dynamics of seed dormancy and germination at high temperature stress is affected by priming and phytohormones in rapeseed (*Brassica napus* L.). Journal of Plant Physiology. 2022;269:153614. https://doi. org/10.1016/j.jplph.2021.153614.

Moraes CE, Lopes JC, Farias CCM, Maciel KS. Qualidade fisiológica de sementes de *Tabernaemontana fuchsiaefolia* A. DC em função do teste de envelhecimento acelerado. Ciência Florestal. 2016;26(1):213-223. doi. https://doi.org/10.5902/1980509821114.

Nonogaki H. Seed germination and dormancy: the classic story, new puzzles, and evolution. Journal of Integrative Plant Biology. 2019;61:541-563. doi. https://doi. org/ 10.1111/jipb.12762.

Oliveira AKM, Souza SA, Souza JS, Carvalho JMB. Temperature and substrate influences on seed germination and seedling formation in *Callisthene fasciculata* Mart. (Vochysiaceae) in the laboratory. Revista Arvore. 2015;39(3):487-495. doi. https://doi. org/10.1590/0100-67622015000300009.

Önol B, Yildirim MU. Breaking seed dormancy and regeneration in *Cannabis sativa* L. International Journal of Agriculture, Environment and Food Sciences. 2021;5(4):709-719. https://doi.org/10.31015/ jaefs.2021.4.32

Pedrini S, Dixon KW. International principles and standards for native seeds in ecological restoration. Restoration Ecology. 2020;28:S286-S303. doi. https://doi. org/10.1111/rec. 13155.

Peng CY, Wu Y, Cai H, Hu YM, Huang WH, Shen YB, Yang H. Methodological and physiological study of seed dormancy release in *Tilia henryana*. Journal of Plant Physiology. 2023;287:154046. doi. https://doi.org/10.1016/j.jplph.2023.154046.

Pinheiro RM, Soares VN, Gadotti GI, Silva EJS da, Almeida AS. Germinative performance of mulungú seeds (*Ormosia* grossa Rudd) after dormancy overcoming. Revista Árvore. 2021;45:e4532. doi. https:// doi.org/10.1590/1806-9088 20210000032.

Porceddu M, Mattana E, Pritchard HW, Bacchetta G. Sequential temperature control of multi-phasic dormancy release and germination of *Paeonia corsica* seeds. Journal of Plant Ecology. 2016;9(4):464-473. doi. https://doi.org/10.1093/jpe/rtv074.

Rosbakh S, Carta A, Fernández-Pascual E, Phartyal SS, Dayrell RLC, Mattana E, Saatkamp A, Vandelook F, Baskin J, Baskin C. Global seed dormancy patterns are driven by macroclimate but not fire regime. New Phytologist. 2023;240:555-564. doi. https://doi.org/10.1111/nph.19173.

Samarasinghe B, Jayasuriya K, Gunaratne A, Senanayaka M, Dixon K. Seed dormancy and germination behavior of tree species in the tropical forest of Sri Lanka. Seed Scientific Research. 2022;32(2):94-103. doi:10.1017/S0960258522000162.

Silva FE da, Pereira MD, Nicolau JPB, Felix FC, Santos GC da S, Araújo JKP de, Almeida R da SA, Bruno R de LA. Morpho-physiological characterization of germination in *Senna siamea*. Revista Ciência Agronômica. 2023;54:e20228425. doi. https:// doi. org/10.5935/1806-6690.20230007.

Silva P, Galastri NA, Mazziero FFF, Gimenez JI. Emergência de plântulas de *Gallesia integrifolia* (Spreng.) Harms (Petiveriaceae) em função do substrato e das condições de armazenamento das sementes. Paubrasilia. 2023;6:e112. doi. https://doi.org/ 10.33447/paubrasilia.2023.0112.

Sousa GG, Sousa HC, Lessa CIN, Goes GF, Freire MHC, Souza MVP, Gomes SP, Schneider F. Production of watermelon seedlings in different substrates under salt stress. Revista Brasileira De Engenharia Agrícola E Ambiental. 2023;27(5):343-351. doi. https://doi.org/10.1590/1807-1929/ agriambi.v27n5p343-351.

Streubel S, Deiber S, Rötzer J, Mosiolek M, Jandrasits K, Dolan L. Meristem dormancy in *Marchantia polymorpha* is regulated by a liverwort-specific miRNA and a clade III SPL gene. Current Biology. 2023;33(4):660-674. doi. https://doi.org/10.1016/j. cub.2022.12.062.

Wolter Filho W, Pinheiro MLB, Rocha AI da. Alcalóides de *Tabernaemontana heterophylla* Vahl. (Apocynaceae). Acta Amazonica 1983;13(2):409-412. doi. https://doi.org/10.1590/1809-43921983132409.

Zhang K, Pan H, Baskin CC, Baskin JM, Xiong Z, Cao W, Yao L, Tang B, Zhang C, Tao J. Epicotyl morphophysiological dormancy in seeds of *Paeonia ostii* (Paeoniaceae): Seasonal temperature regulation of germination phenology. Environmental and Experimental Botany. 2022;194:104742. doi. https://doi. org/10.1016/j.envexpbot.202 1.104742.